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METFORMIN AND STATIN'S EFFECT ON PROSTATE CANCER RISK IN DIABETICS
WITH BENIGN PROSTATIC HYPERPLASIA

A Thesis Presented to
The Faculty of the School of Medicine
Yale University

In Candidacy for the Degree of
Master of Medical Science

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Abstract

Prostate cancer, a leading cause of cancer mortality, is associated with benign prostatic hyperplasia. Additionally, individuals with type 2 diabetes have a diminished prostate cancer risk. Metformin and statins are routinely used in the management of diabetes; however, it is unclear whether the combination of metformin and statins can reduce the incidence of prostate cancer in men with a concurrent diagnosis of type 2 diabetes and benign prostatic hyperplasia.

This study will determine whether combination treatment of metformin and statins lowers the incidence of prostate cancer in patients with benign prostatic hyperplasia and type 2 diabetes. Using a retrospective cohort study, we will compare the incidence of prostate cancer among type 2 diabetics with benign prostatic hyperplasia using both metformin and statins against cohorts using metformin only, statin only, and neither metformin nor statin. These results will elucidate metformin and statin's role as preventative cancer modalities.

Chapter 1: Introduction

1.1 Background

Prostate cancer is the second leading cause of cancer-related mortality amongst males in the United States.¹ Over one million new cases of prostate cancer were reported worldwide in 2018, with North America maintaining the second highest prostate cancer incidence rates in the world.¹ The prevalence of prostate cancer increases with age and rises significantly after the age of 50, paralleling the increase in individuals within the elderly population.² Therefore, the global incidence of prostate cancer diagnoses over the past 20 years has gradually increased, partly due to a surge in the number of individuals in the aging population.³ While appropriate implementation of early detection modalities have led to a statistically significant reduction in the mortality of prostate cancer,⁴ the mortality rate amongst those with prostate cancer is still elevated, as current estimates project that over 30,000 men are expected to die from this disease each year.⁵ Although there have been advancements in the ability to diagnose and treat prostate cancer, early recurrence is still common, with up to 50% of patients with localized prostate cancer experiencing recurrence within 10 years.⁶ Therefore, despite improvements amongst prostate cancer treatments, the most effective therapeutic approach is primary prevention of prostate cancer.

Interestingly, men with type 2 diabetes mellitus (T2DM) are less likely to develop prostate cancer when compared to patients without T2DM.⁷ However, the mortality rate amongst those with T2DM and prostate cancer is higher when compared against non-diabetics, particularly amongst those with inadequately controlled blood sugars.⁸ Individuals with T2DM have a poorer prognosis for prostate cancer due to various mechanisms, including hyperglycemia induced tumor progression⁹ and increased rates of radiotherapy treatment failure.¹⁰ Alternatively,

controlling blood sugar levels in those with T2DM may provide an opportunity to reduce the possibility of hyperglycemia induced tumor progression. Therefore, treatment of T2DM with metformin, an oral hypoglycemic agent, may improve prostate cancer outcomes for those with T2DM.¹¹⁻¹³ Further, commonly utilized medications for treating T2DM, specifically metformin and statins, may be responsible for the reduced incidence of prostate cancer diagnoses amongst those with T2DM.

Metformin is a first line medication for the treatment of T2DM, and has demonstrated anti-neoplastic effects in a variety of solid tumors, such as colon cancer, pancreatic cancer, and breast cancer.¹⁴⁻¹⁶ Further, meta-analyses and systemic reviews have determined that metformin reduces the incidence of prostate cancer and improves prostate cancer outcomes,^{17,18} although other studies were unable to substantiate these findings.¹⁹ The discrepancies in the literature may be due to several factors, including differing populations studied, inclusion criteria variation, dissimilar treatment durations, and different markers of prostate cancer development. Therefore, the true influence of metformin on the incidence of prostate cancer amongst those with T2DM requires further investigation.

Despite continued exploration into the clinical anti-cancer action of metformin, the underlying biophysiological mechanisms of metformin's antineoplastic effects are better understood. Laboratory studies have elucidated metformin's role in genetic,²⁰ cell growth,^{13,21-28} and cell division²⁹⁻³¹ mechanisms that contribute to metformin's anti-neoplastic action on cancerous cells. However, the clinical relationship between metformin and incidence of prostate cancer necessitates further elucidation. Additionally, a potential synergistic effect of metformin and statin therapies may illuminate the risk reduction in prostate cancer found amongst metformin users.³²

Statins are widely utilized amongst diabetic populations due to their ability to reduce the risk for cardiovascular diseases. As many individuals with T2DM have significant risk factors for future cardiovascular diseases, the American Diabetes Association recommends that statin therapy for individuals with T2DM should be based on the inherent cardiovascular risk that accompanies diabetes.³³ Further, several studies have highlighted the potential role of statins in the chemoprevention of many cancer types.^{34,35} While statins have been associated with a reduction in the incidence of several cancers, the most promising data pertains to prostate cancer risk.^{36,37} For instance, it has been suggested that statins are partially responsible for the steep decline in the prostate cancer death rate in the US during the past 15 years.³⁸

The underlying physiological reasoning behind statin's ability to lower the risk of prostate cancer has been explored in laboratory studies, which found that statins may prevent the development of prostate cancer through inhibition of proliferative signals, sensitizing potentially malignant cells to programmed cell death, minimizing inflammation, reducing angiogenesis, and impeding invasiveness by blocking adhesion molecules.³⁹

Despite the evidence supporting statin use to lower the incidence of prostate cancer, not all studies agree on the potential anti-neoplastic effect of statins. Analyses from the Prostate Cancer Prevention Trial (PCPT) and Reduction by Dutasteride of Prostate Cancer Events (REDUCE) were unable to determine a significant reduction of prostate cancer incidence amongst statin users.^{40,41} While, individual case-control and cohort studies have conflicting findings, a 2012 meta-analysis reported a significant reduction in the risk of prostate cancer in statin users in comparison with nonusers.⁴² However, several recent studies have failed to find an association between statin use and a reduced risk of advanced prostate cancer. Interestingly, a study with the greatest number of subjects compared to all other related studies to date reported a

significantly reduced risk of advanced prostate cancer in statin users,³⁶ which is in line with other findings.³⁷ However, given the heterogeneity amongst the literature, sufficient data is lacking to support the use of statins for the primary prevention of prostate cancer. Further, none of the previously mentioned studies examined the combination of metformin and statins on prostate cancer risk. Therefore, it is imperative to examine potential synergist effects of metformin and statins in lowering the incidence of prostate cancer.

When evaluating the development of prostate cancer amongst a population, utilizing subjects at an inherently increased risk can lead to stronger conclusions with shorter study time frames, which is of particular interest when completing observational studies. Therefore, as benign prostate hyperplasia (BPH) is associated with an increased incidence of prostate cancer,⁴³ it is important to study this population when examining metformin and statin's effect on prostate cancer. Much like prostate cancer, the increasing prevalence of BPH parallels with the rise in the aging population.⁴⁴ There is a theorized relationship between BPH and the development of prostate cancer, as these prostatic diseases share common pathophysiological driving factors.⁴⁵ Many prostate cancer patients have a previous diagnosis of BPH,⁴³ and those with prostate cancer have a high prevalence of symptoms associated with BPH.⁴⁶ Despite this relation between BPH and prostate cancer, research suggests that BPH is not a known risk factor for prostate cancer.⁴⁷ However, in a 2016 meta-analysis of 19 studies, all studies showed an increased prostate cancer diagnoses amongst those with BPH.⁴³ Therefore, a diagnosis of BPH increases the incidence of prostate cancer diagnoses among this population due to increased screening and incidental cancer diagnoses, thereby allowing for a suitable study population, despite the lack of evidence for BPH serving as an independent risk factor for prostate cancer.

The incidence of BPH is influenced by various systemic conditions. For instance, the risk of developing BPH is increased amongst diabetic populations because diabetes mellitus is associated with an increased sympathetic tone, heightened prostate growth by insulin related trophic factors, alterations in sex steroid hormone expression, and an induction of systemic inflammation and oxidative stress.⁴⁸ Therefore, there are a significant number of individuals with a concurrent diagnosis of T2DM and BPH, as these two diagnoses frequently coexist.

The previously mentioned differences in the findings for potential the risk reduction of prostate cancer in both metformin and statins, along with the influence of BPH on diagnosing prostate cancer, highlight the necessity to further examine the potential relationship between metformin, statins, BPH, and prostate cancer. As older men with BPH may incur a heightened baseline incidence of prostatic carcinomas, it is imperative to determine treatment approaches to lower this population's risk of developing prostate cancer.

1.2 Statement of the problem

Although both metformin and statins have been associated with a reduced risk of prostate cancer incidence, the relationship between metformin, statin, BPH, and prostate cancer requires further examination. A study conducted in Taiwan suggested that metformin may be associated with a reduced risk of prostate cancer diagnosis for men with both BPH and T2DM.⁴⁹ However, no studies have examined the influence of both metformin and statins in the risk of prostate cancer among individuals with BPH and T2DM.

Examining the relationship between anti-diabetic therapy and prostate cancer through BPH can help better understand the implications of statin and metformin treatments amongst populations at increased incidence of prostate cancer diagnoses. This approach has been successfully implemented by one study,⁴⁹ although statin use was not explored. Further, this experiment did not control for various BPH medications, which reduces the internal validity of

their conclusions.⁴⁹ 5-alpha reductase inhibitors, a common medication to control BPH symptoms, may lower the incidence of prostate cancer in BPH individuals.⁵⁰ Therefore, targeted medications for controlling BPH symptoms should be stratified between subjects using a 5-alpha reductase inhibitor and those utilizing other common BPH treatment modalities, such as alpha blockers.

Metformin and statins may have broader clinical applications as medications for cancer prevention due to their low cost and well-known safety profile.^{51,52} The use of metformin and statins as preventive cancer treatments may expand outside of the diabetic population. For instance, a clinical trial of metformin use in nondiabetic patients with prostate cancer resulted in 36% of the subjects with progression-free prostate cancer at 12 weeks.⁵³ Therefore, metformin and statins have the potential to be widely utilized as a treatment modality to reduce the risk of developing prostate cancer.

1.3 Goals and objectives

The objective of this study is to determine the relationship between metformin, statins, and prostate cancer incidence among individuals with BPH and T2DM. This research is critical because the potential synergistic effects of metformin and statins on the risk of prostate cancer amongst populations with BPH has not been evaluated.

The goal for this study is to examine if statins or metformin may be used in the primary prevention of cancers, and to further explore the anti-neoplastic effects of statins and metformin.

1.4 Hypothesis

Individuals aged 50 and above with type 2 diabetes mellitus and benign prostatic hyperplasia who are treated with metformin and statins between the years 2009 to 2019 will have a statistically significant difference in the incidence of prostate cancer when compared to those using metformin only, statin only, or neither metformin nor statins.

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Chapter 2: Literature Review

2.1 Introduction and Methodology

The literature review was performed between July 2020 and January 2021 using the following databases: PubMed, Cochrane library, and Scopus. Searches were initially performed using the following MeSH terms: *Metformin*, *Statin*, *Benign Prostatic Hyperplasia*, and *Prostate cancer*. Alternative search terms for metformin included: *oral hypoglycemic agent*, and *anti-diabetic medication*. Alternative search terms for statin included: *HMG-CoA Reductase Inhibitor*, *LDL lowering medication*, *lipid lowering medication*, *cholesterol lowering medication*, and various statin brand and generic names. Alternative search terms for benign prostatic hyperplasia included: *BPH*, and *benign prostatic hypertrophy*. Alternative search terms for prostate cancer included: *prostatic carcinoma*, *prostate adenocarcinoma*, and *prostate malignancy*. After completion of the primary literature search, additional references were extracted from the references of previously utilized papers when appropriate. Studies examined included clinical studies, laboratory studies, systemic reviews, and meta-analyses. After determining article relevance, they were analyzed for inclusion in the literature review chapter.

2.2 Review of studies about relationship of metformin, statins, and prostate cancer

Numerous experiments examining the effect of metformin and statins on prostate cancer development and progression have arrived at diverse conclusions. Additionally, various observational studies have determined the impact of metformin or statin use on the incidence of prostate cancer. Interestingly, these investigations have also resulted in inconsistent conclusions regarding the influence of either metformin or statins on prostate cancer incidence. Despite the different conclusions, there is considerable evidence that metformin and statins can lower the incidence of prostate cancer amongst diabetics with BPH.

2.2.1 Review of the association between metformin and prostate cancer incidence

Several observational studies have investigated a potential association between metformin and the incidence of prostate cancer. Three studies have demonstrated a reduction in the incidence of prostate cancer amongst diabetic metformin users.¹⁻³ When examining the proposed association between metformin and prostate cancer risk, it is necessary to control for other medications that are commonly utilized amongst diabetic populations. To address this, Ruiter et al. compared the risk of prostate cancer in diabetic patients beginning metformin therapy against those starting other classes of diabetes medications. They found that metformin is associated with a lower risk of prostate cancer, and has a stronger reduction in prostate cancer development when compared to other hypoglycemic agents (Hazard Ratio (HR) 0.92; 95% CI 0.88-0.97; $P < 0.05$).¹ This highlights that when controlling for non-metformin diabetes treatments, there is still a significant reduction of prostate cancer risk amongst metformin users.

As our proposed study population will utilize subjects who have an increased baseline incidence rate for prostate cancer diagnoses given their age and BPH diagnosis, it is critical to examine metformin's role in preventing prostate cancer development amongst populations that have similar heightened rates of prostate cancer diagnoses. One study examined subjects with high grade prostatic intraepithelial neoplasia, a high-risk pre-cancerous state, and determined that metformin is linked to a decreased incidence of prostate cancer compared to non-users ($p < 0.001$).² Comparably, a retrospective cohort study examined diabetics with BPH and determined a reduced risk of prostate cancer in subjects with BPH who used metformin compared to the non-metformin cohort (adjusted HR (aHR) 0.69; 95% CI 0.49-0.96; $P = 0.0298$).³ This evidence suggests that metformin may lower the risk of prostate cancer amongst individuals that are similar to our studied population.

Nevertheless, other studies have failed to report a statistically significant association between metformin and prostate cancer incidence.⁴⁻⁶ A recent meta-analysis concluded that while metformin may be protective against prostate cancer in certain subgroups, such as metformin compared against sulfonylureas, the pooled analysis revealed no statistical significance for metformin use and reduced prostate cancer risk (case-control HR 0.97; 95% CI 0.84-1.12; cohort HR 0.94; 95% CI 0.79-1.12; $P < 0.05$).⁷ However, some of these studies that failed to find an association of metformin and prostate cancer contain inherent drawbacks and design flaws.

For instance, a nested case-control study concluded that metformin use did not decrease the risk of prostate cancer in diabetic patients utilizing an oral hypoglycemic agent (adjusted relative risk (aRR) 1.23; 95% CI 0.99-1.52; $P < 0.05$).⁴ However, this study did not independently analyze metformin users against those utilizing other oral hypoglycemic agents. As previously discussed, there is evidence that metformin lowers the incidence of prostate cancer when compared against other hypoglycemic agents.¹ Therefore, this study's conclusions are influenced by other agents that can influence the studied risk of prostate cancer development.

Other studies may have failed to find an association between metformin and a reduced incidence of prostate cancer due to the underlying characteristics of prostate cancer treatment modalities. For instance, a prospective cohort study demonstrated that the use of metformin is associated with an increased risk of high-grade prostate cancer in men who have previously undergone a radical prostatectomy (odds ratio (OR) 3.11; 95% CI 1.16-8.33; $P = 0.012$).⁵ Interestingly, all of the subjects in this study were treated surgically, unlike the subjects in a similar study that determined a reduction in prostate cancer mortality amongst metformin users, in which only 7.7% of the subjects were managed surgically (aHR 0.76; 95% CI 0.64-0.89; $P < 0.05$).⁸ Conversely, a retrospective cohort study found no association amongst metformin and

incidence of high-grade prostate cancer recurrence in patients treated with a radical prostatectomy (HR 0.84; 95% CI 0.58-1.22; P=0.36).⁶ When compared to the subjects of the previous study,⁸ the subjects of this study⁶ had significantly lower rates of positive surgical margins, which is a risk factor for high grade prostate cancer.⁸ Therefore, the association between metformin and prostate cancer may significantly differ based upon the treatment modalities of prostate conditions in the subjects.

2.2.2 Review of the association between metformin and progression of prostate cancer

Understanding metformin's ability to inhibit the progression of prostate cancer can further delineate its proposed clinical anti-neoplastic effects. Regarding prostate cancer specific outcomes, studies have examined metformin in relation to biochemical recurrence, castration resistant prostate cancer, distant metastases, and prostate cancer specific mortality.⁹⁻¹⁴ In three observational studies, the use of metformin was associated with a decreased risk of prostate cancer progression.^{8,10,15} However, three other studies reported no reduction in prostate cancer progression or mortality.^{9,12,13} One such study utilized a large cohort of 885 patients, and the researchers concluded a lack of effect of metformin on improving prostate cancer outcomes (HR 1.16; 95% CI 0.73-1.86; P=0.53).¹⁴ However, this result is inconsistent with an examination of prostate cancer patients who underwent external beam radiation therapy, which determined that metformin use was associated with a significantly reduced risk of prostate cancer progression (HR 5.15; 95% CI 1.53-17.35; P=0.008).¹⁰ These inconsistencies can be attributed to various factors, including time biases and differences amongst subject demographics.

Some studies that found a decreased risk of prostate cancer progression amongst metformin users failed to analyze metformin-use as a time-dependent variable. Failing to adjust for metformin use as a time dependent variable creates an immortal time bias, which has been demonstrated to exaggerate the benefits of metformin on prostate cancer outcomes in

observational studies.¹⁶ Therefore, this bias amongst studies that found a reduction of prostate cancer progression in metformin user may partly explain the inconsistencies in the literature.

This bias will be further examined in the “confounders” section of this paper.

A recent meta-analysis determined that a majority of the inconsistencies in the literature surrounding metformin and prostate cancer outcomes were due to obesity and prostate specific antigen (PSA) adjustments in statistical models.⁷ Differences among an individual’s BMI can affect the results as obesity influences the rate of hyperglycemic progression of prostate cancer.¹⁷ Additionally, PSA significantly contributes to the incidence of prostate cancer diagnoses, which is further discussed in the “confounders” section of this chapter. Therefore, differences amongst subject’s obesity and PSA screening levels can explain the inconsistencies between these studies.

These findings highlight the discrepancies amongst observational studies examining the effect of metformin on prostate cancer development and progression. Despite these inconsistencies, there is strong evidence supporting an association between metformin and a reduction of prostate cancer progression. Therefore, this clinical evidence allows for the deduction that metformin may similarly inhibit the development of prostate cancer, as reflected by a reduction in the incidence of prostate cancer among metformin users.

2.2.3 Review of the association between statins and prostate cancer incidence

Similar to metformin, the evidence of an association between statins and the risk of prostate cancer is ambiguous, with relative risks varying between 0.26 to 2.94.¹⁸ While some studies demonstrate a protective effect of statins on the risk of prostate cancer,¹⁹⁻²⁷ others conclude that statins are unable to reduce the incidence of prostate cancer.²⁸⁻³²

A large case control study of 4,204 men undergoing prostate biopsies at consistent 2-year intervals reported a 8% reduced risk of total prostate cancer in statin users in comparison with nonusers (RR 0.92; 95% CI 0.85-0.98; P<0.05).³³ Interestingly, frequent screenings for prostate

cancer is associated with increased incidence for prostate cancer, as such is the theorized association between BPH and prostate cancer.³⁴ However, the above-mentioned study determined a reduction amongst prostate cancer diagnoses in statin users despite frequent screening, which strengthens this proposed association. Similarly, a large population-based case control study, which included over 40,000 patients with any stage of prostate cancer and over 200,000 controls, reported a significant 6% reduction in the risk of prostate cancer amongst statin users (OR 0.94; 95% CI 0.91-0.97; $P < 0.05$).³⁵ This conclusion has been replicated in other study designs, as a retrospective study from a cohort of over 55,000 men in the Veteran's Association found that statin users were 31% less likely to be diagnosed with prostate cancer when compared to men who did not use statins (HR 0.69; 95% CI 0.52-0.90; $P < 0.05$).³⁶ Other studies with smaller study populations found similar associations between statin use and a reduction in prostate cancer risk.^{24,31} However, some of these studies did not stratify statin users based upon the two major categories of statins: hydrophobic and hydrophilic statins. This stratification is important as there is a proposed difference in the effect of these different statin types on the risk of prostate cancer.

A retrospective cohort study examining 37,645 men taking statins found a 74% reduced risk of total prostate cancer in long term hydrophobic statin users, defined as greater than 5 years of statin use, in comparison with nonusers (HR 0.48; 95% CI 0.37-0.56; $P < 0.05$).³⁷ Notably, this same study found that hydrophilic statins had no association with a reduction in the risk of prostate cancer when compared to hydrophobic statins (HR 0.92; 95% CI 0.75-1.17; $P < 0.05$).³⁷ Therefore, failing to adjust for the type of statin used may contribute to the inconsistencies found amongst the literature. This association amongst statin type and prostate cancer risk will be further detailed in this chapter.

Despite the findings of statin use and a reduced risk of prostate cancer, other observational studies have failed to find an association amongst statin users and prostate cancer risk. For instance, a secondary analysis of a randomized trial in men with a previously negative prostate biopsy who underwent repeat biopsies at 2 and 4 years found no reduction in prostate cancer risk and statin use (multivariate OR 1.05; 95% CI 0.89-1.24; P=0.54).³⁰ A similar study found that among men who were screened for prostate cancer annually despite PSA levels, statins did not protect against prostate cancer (HR 1.27, 95% CI 0.85-1.90; P<0.05).²⁹ However, both of these studies screened subjects for prostate cancer more aggressively than current medical recommendations.³⁸ Therefore, these results may be falsely skewed against a potential prostate cancer risk reduction as frequent screening may diagnose cancers that would otherwise be undetected in this population.³⁹

Additionally, a case control study in a Canadian population found that there was no association amongst statin use and prostate cancer incidence (OR 0.96; 95% CI 0.90-1.03; P<0.05).³² However, this study did not control for PSA testing amongst the groups, which may influence the results as statin users undergo PSA testing more frequently when compared to non-statin groups.⁴⁰ Similarly, a cohort study among elderly US men have found no association between statin use and prostate cancer risk (OR 1.24; 95% CI 0.98-1.57; P=0.07),²⁸ however this study lacked information on the PSA screening rates of the cohorts. These discrepancies amongst the observational studies may be related to methodological issues, including differences in sample size, assessment of statin use, PSA testing activity, and other confounding factors. Therefore, the findings of a reduced prostate cancer risk when controlling for some of these variables, such as PSA screening and statin type, suggest that there is a true reduction in the incidence of prostate cancer amongst statin users.

2.2.4 Review of the association between type of statin and prostate cancer incidence

The major classifications of statins are divided between hydrophilic and hydrophobic statins. Interestingly, the risk of prostate cancer has been theorized to vary depending on the type of statin used. Hydrophobic statins are easily taken up by non-hepatic tissues, such as the prostate⁴¹ and therefore are hypothesized to have a greater influence on the prostate when compared to hydrophilic statins. This has been evidenced through an observational study, which found that hydrophobic statins reduce the hazard of incident prostate cancer, whereas hydrophilic statins as a group show no association with incident prostate cancer (HR 0.921 95% CI 0.75-1.17; $P < 0.05$).³⁷ However, this is not a consistent finding in the literature, partly because the number of men taking hydrophilic statins in several studies were low.³⁵

Interestingly, these results are contrasted by a recent study which determined that hydrophilic statins showed a greater association with decreased prostate cancer diagnosis rates ($P < 0.05$).²³ However, this study had dissimilar numbers of medication users within their groups, as two hydrophobic statins- atorvastatin and rosuvastatin- had the lowest number of users, but showed a protective effect on prostate cancer. Similar inconsistencies between statin type and association with prostate cancer were demonstrated in the Finnish prostate screening trial.⁴²

Despite these findings, the differences between hydrophobic and hydrophilic statins are not consistently appreciated as a case control study concluded that risk estimates did not differ substantially by type of statin used.³⁵ This result has been corroborated by another study,²⁸ however this study was limited by a small sample size, self-reported statin usage, and lacked data on duration or dose of statin usage.

These findings highlight that the mechanism of action in reducing cancer cell proliferation is not solely dependent on a statin's ability to reduce cholesterol, as this would cause a uniform reduction in cancer incidence independent of statin type. Thus, a number of

different mechanisms are working simultaneously, and not all statin types trigger these mechanisms.³⁷ In most other epidemiological studies to date, analyses were not performed for individual statin groups; if the effects of statins on incident prostate cancer varies between statin type, then variations in statin usage may explain why some studies have not found an overall association between statin use and incident prostate cancer.

2.3 Review of studies to identify possible confounding variables

2.3.1 *Metformin duration*

The duration of metformin and statin usage is a considerable factor when examining their influence in the development of prostate cancer. The effect of the duration of metformin use on prostate cancer prevalence was examined through a retrospective cohort study, which concluded that a longer duration (>2 years) of metformin use was associated with a lower incidence of prostate cancer amongst subjects with high-grade prostatic intraepithelial neoplasia ($P < 0.001$).² Another study determined that metformin use <1.5 years was not associated with a prostate cancer risk reduction, but durations >3 years were associated with a decreased prostate cancer incidence (aOR 0.84; 95% CI 0.74-0.96; $P < 0.05$).⁴³ Thus, a multiple-year duration of metformin treatment further enhances the medication's ability to exert anti-neoplastic effects, thereby highlighting the need to consider the duration of metformin usage when examining the association amongst metformin treatment and prostate cancer prevalence. However, inconsistencies remain in the literature of a potential duration dependent effect of metformin on prostate cancer incidence, as a 2020 meta-analysis found that while there was a slightly protective effect of metformin compared to sulfonylureas, there was no significant association amongst cohort studies that had a longer duration (over 5 years) of metformin use when compared against those using shorter durations (under 5 years) (HR 1.02; 95% CI 0.87-1.20; $P = 0.813$).⁷ Therefore, while the duration of metformin use is not consistently identified as a

significant bias in this study setting, it is still crucial to factor for the duration of metformin use when assessing prostate cancer incidence.

2.3.2 *Metformin dose*

The dose of metformin may contribute to the efficacy of its anti-neoplastic potential, thereby acting as a confounding variable amongst observational studies. A systemic review recently concluded that several cellular studies have demonstrated a dose-dependent effect of metformin for reducing the risk of developing various cancers, however these studies utilize supra-physiologic doses.⁴⁴ To date, it remains to be evaluated clinically if the conventional prescribed doses of metformin therapy for the treatment of type 2 diabetes can influence the development of prostate cancer.⁴⁴

2.3.3 *Statin duration*

It is critical to assess the duration of statin use when determining the association of statins and the incidence of prostate cancer. The various durations of statin usage in studies that have analyzed the association of statin use and prostate cancer contribute to the discrepancies between these studies. Many cohort and case control studies are limited by exposure data being confined to the recent past, limited information on dose, timing and duration of statin use, and by the possibility of uncontrolled detection and recall biases.²¹ However, a study that examined the influence of detection bias in the association of statin and prostate cancer risk determined that detection bias is unlikely to explain the reported inverse association between statins and advanced prostate cancer, however it may explain the positive association for total prostate cancer that has been reported in some studies of statin use.⁴⁵ Therefore, this conclusion assumes an increased inverse association between the duration of statin use and risk of prostate cancer in studies that have unintentionally introduced a detection bias. Hence, controlling for the duration

of statin use amongst subjects is needed to improve the strength of the potential association between statin use and prostate cancer risk.

2.3.4 BMI and insulin resistance

Another important confounding factor is a subject's body mass index and associated insulin resistance. Experimental studies have concluded that body mass index and insulin resistance independently influence cancer prognosis through tumor cell proliferation, invasion, and metastasis.⁴⁶ While findings on obesity and prostate cancer development have been mixed, it has been demonstrated that obesity is consistently associated with a higher risk of aggressive prostate cancer.⁴⁷ Additionally, meta-regression analyses found that obesity in statistical models may be one of the main contributors of heterogeneity amongst studies on the effect of metformin on prostate cancer development.⁷ Therefore, it is imperative to control for body mass index when examining metformin cohorts against non-metformin cohorts. However, while controlling for BMI is critical, it does not directly control for insulin resistance amongst patients. Controlling for insulin resistance is critical because metformin's proposed antitumor effect stem from both direct biophysiological and indirect insulin lowering factors.⁴⁸ Therefore, studies should consider patient insulin usage at baseline, and may benefit from including a cohort of non-diabetic patients for comparison.⁴⁹ However, this factor introduces study design challenges as metformin is currently only indicated for diabetic populations. Therefore, one must consider the rationale and potential consequences when designing a study in which non-diabetic patients are administered an oral hypoglycemic agent.

2.3.5 Baseline fasting glucose level

A confounding factor worth considering is a subject's ability to manage their diabetes. Sub-standard glycemic control may enhance the growth of cancerous prostate cells as hyperglycemic states stimulate cancer development and proliferation.⁵⁰ Additionally, poor

control of a subject's diabetes may stem from sub-standard metformin therapy, which would inaccurately under treat those in the exposure group, leading to bias towards the null hypothesis. Thus, standardization of glycemic control amongst the cohorts would prevent the potential influence of this confounder.

2.3.6 Mean duration of diabetes

Another important confounding factor is the mean duration of diabetes amongst the cohorts. As diabetes may offer a protective effect on prostate cancer development,⁵¹ a longer duration of diabetes may correlate with additional protective effects against prostate cancer growth. Therefore, a cohort with an average longer duration of diabetes diagnoses would be at a reduced risk to develop prostate cancer. It is crucial to ensure that all cohorts have a similar mean duration of diabetes to eliminate this bias.

2.3.7 Baseline cholesterol levels

An additional potential confounder in the association of statins and prostate cancer incidence is baseline cholesterol levels in statin users, as cholesterol levels may alter growth of cancerous prostate cells. Baseline cholesterol levels may confound results as effective adherence to statin use would significantly reduce low-density lipoprotein cholesterol (LDL).⁵² However, a study that analyzed the relation of cholesterol levels and prostate cancer incidence found that the association between statin use and prostate cancer risk did not significantly differ between men with serum cholesterol under 200 against those over 200 at trial entry (HR 1.03; 95% CI 0.82-1.30; $P < 0.05$).²⁹ Therefore, while baseline cholesterol levels may be a confounder in the association of statin and prostate cancer incidence, it has not been shown to significantly alter the risk of developing prostate cancer.

2.3.8 PSA levels

Baseline PSA levels may be another potential confounding factor amongst the proposed cohorts. Statins are associated with lower PSA levels as 1 year of statin use can reduce PSA

levels by 4.1% ($P<0.05$).³⁰ While some studies on the association of statins and prostate cancer adjusted for PSA levels, others did not. It is crucial to control for PSA levels, as prostate cancer diagnosis is greatly influenced by PSA levels, which is in turn influenced by statin use.³⁰ As PSA is a primary method for prostate cancer screening, this confounds any associations between statins and the risk of being diagnosed with prostate cancer. A study that examined the association between statins and prostate cancer in mostly PSA independent diagnoses found no association amongst statin use and risk of prostate cancer (OR 1.05; 95% CI 0.89-1.24; $P=0.54$),³⁰ which suggests that a reduced amount of PSA screening in statin users may falsely lower the assumed risk of prostate cancer amongst statin users. However, a 4% decline in PSA levels³⁰ may not reduce the number of biopsies enough to sufficiently explain the lower incidence prostate cancer diagnoses found in statin users.^{20,23,26} Therefore, one must examine PSA levels amongst the cohorts when analyzing the effect of medications on prostate cancer risk.

2.3.9 Detection bias

Detection in this literature refers to how measures of detecting of prostate cancer bias the determined association.²⁹ Detection bias in prostate cancer studies could result from the possible influence of statins on serum PSA, and from the tendency for men seeking medical care to receive regular screening for both elevated cholesterol and PSA.⁴⁵ This is noted by an increased amount of PSA screening among statin users, as those utilizing statins may be more inclined to undergo prostate cancer screening. Although a 2012 meta-analysis reported a relative risk of prostate cancer of 0.93 (95% CI; 0.87-0.99; $P<0.05$) amongst statin users,⁵³ a more recent review⁵⁴ highlighted significant heterogeneity between published studies, many of which relied on questionnaires for exposure data and lacked adjustment for prostate cancer screening.¹⁸

A study that controlled for PSA screening found no association amongst prostate cancer and statin use (OR 0.96; 95% CI 0.90-1.03; $P<0.05$).³² Similarly, a study reduced detection bias

by examining prostate cancer incidence amongst statin users who were regularly screened for prostate cancer through PSA testing.²⁹ This study found no association amongst statin use and prostate cancer (HR 1.03; 95% CI 0.82-1.30; $P < 0.05$).²⁹ However, this study had no information on statin use. These results contrast a study with a long follow up period of over 90,000 person-years with recurrent PSA testing, which found that one year of statin use was associated with a decreased risk prostate cancer (aHR 0.85; 95% CI 0.42-0.69; $P < 0.05$).²⁷ This finding indicates that detection bias may not fully explain the association of prostate cancer risk and statin use.²⁷

A cohort study reported a 25% reduction in prostate cancer risk amongst statin users participating in the PSA screening arm of a prostate cancer screening trial in which PSA tests were available for all participants.⁴² This understanding is furthered by a cohort study in which PSA measures were only available for symptomatic subjects, which mimics the typical PSA screening process, and found that statin use was associated with a decreased incidence of prostate cancer (HR 0.82; 95% CI 0.74-0.93; $P < 0.05$).³⁷ Therefore, routine PSA screening may falsely increase the risk estimates for prostate cancer amongst statin users. Therefore, excessive PSA screening may cause detection bias that influences a false positive association between use of statin and increased risk of prostate cancer. However, a 2017 review concluded that while increased PSA screening may bias some findings, they are unlikely to fully explain the inverse association between statin use and prostate cancer risk.⁵⁴ Therefore, proper adjustment of PSA screening amount amongst the cohorts is necessary to control for this potential confounder.

2.3.10 Time-related bias

When completing an examination of medication use, it is critical to avoid introducing an immortal time bias, time-window bias, or time-lag bias. All three of these time related biases may greatly exaggerate the observed benefits of metformin or statins in reducing the risk prostate cancer development.¹⁶ Of the time-related biases, the most important to avoid in our study is an

immortal time bias, as this may be introduced with a time-fixed cohort analysis.¹⁶ This bias is introduced when participants are assigned to an exposure group prior to receiving the exposure (i.e. classification of medication use prior to filling a prescription). This bias has been suggested to contribute to the heterogeneity found amongst the literature of medication use and prostate cancer risk.¹⁶ An immortal time bias causes the resulting analyses to produce apparent reductions in risk that are created artificially by the misclassification of metformin exposure. A recent meta-analysis of confounding factors in observational studies of metformin and prostate cancer risk determined that studies that accounted for time-related biases found that metformin was not associated with the risk of cancer incidence because the presence of these biases may exaggerate the protective effects of metformin.⁷ However, immortal time bias can be avoided by classifying medication use as a time-dependent variable.⁵⁵ Therefore, when designing this observational study, it is crucial to design and analyze this study with careful consideration of time biases to best understand the true association of metformin and prostate cancer incidence.

Another time-related bias found in the literature is a misclassification of exposure timing. This bias is found when subjects utilize the exposure medication for long periods of time prior to the index date. To avoid this bias, a sub analysis can be performed in which only new users of metformin are analyzed, as identified by men who did not have any prior use before the baseline cohort entry.⁵⁵ Thus, when assessing the criteria for metformin use, it is imperative to assess metformin as a time dependent variable, and to quantify the minimum use to be long enough to appreciate metformin's potential anti-neoplastic effects.

2.4 Review of relevant methodology

2.4.1 Variable measurements

Several different methods have been utilized to examine the effect of medication on prostate cancer risk. Studies that have examined the incidence of prostate cancer have defined a

prostate cancer event as having an ICD-9 diagnosis of 185.xx from a primary or secondary diagnosis code.⁵⁶ Amongst studies on the effects of metformin or statins on developing prostate cancer, most examine the incidence rate of the first prostate cancer diagnosis during the study period.^{4,56} Thus, the dependent variable is the amount of new prostate cancer diagnoses among the subjects,^{1-4,7,20-26,56} with an occasional dependent variable as the time interval between initiation of the medication to the first prostate cancer diagnosis observed during the study period.^{55,56}

2.4.2 Study design

The ideal study design to examine the relationship of metformin and statins on the risk of developing prostate cancer in patients with BPH would be a randomized control trial. However, this study design is not feasible to perform in a two-year time frame, as it would not allow for adequate duration needed for the development of potential outcomes within each group.

Therefore, a retrospective cohort or case-control study are the best design options, as these study designs would eliminate the need for following participants for an extended time until they develop prostate cancer. Additionally, given the difficulties with developing a randomized control trial in cancer prevention studies, no previous studies have completed a randomized control trial to explicitly study metformin or statin's association with prostate cancer incidence. Therefore, when examining the effects of metformin and statins on the incidence of prostate cancer in men with benign prostatic hyperplasia, studies commonly utilized either a cohort analysis^{2,3,11,19,22,26-29,37,55-57} or case-control analysis.^{4,12,21,32,35,58,59} One study designed a retrospective cohort study but analyzed the data in a nested case-control design.¹¹ In this study, a nested case-control approach was chosen as the primary analysis rather than the cohort design because the effects of cumulative exposure could not be tested in the cohort design.¹¹ However, given the timing limitations of the current study, a case control design would not allow sufficient

time for an accurate representation of the population's risk for developing prostate cancer, as this cancer is typically slow-growing, and therefore takes several years to arrive at the proper diagnosis.⁶⁰ Therefore, a retrospective cohort analysis would offer a proper duration to examine the subject's risk of developing prostate cancer. Additionally, selecting the cohort in a retrospective study reduces the burden of following participants for an extended amount of time, as participants already have the outcome of interest.

Further, the only study that has examined the effect of both metformin and statins on the incidence of prostate cancer assigned subjects into four medication groups (metformin/no statin, metformin + statin, no metformin/no statin, no metformin + statin).⁵⁶ This design allows for examination of all potential statin and metformin combinations, and therefore will be utilized in our proposed study.

2.4.3 Defining Metformin Use

It is important to understand how metformin use is defined in previous studies to best understand how to put previous findings into context, and to adequately define metformin use in our study. There is substantial heterogeneity amongst metformin use in the literature. One study classified sufficient metformin exposure as a minimum of 180 days of usage, as defined by the amount of prescriptions filed.⁵⁶ However, those that only used metformin for the minimum duration in this study may not have been able to appreciate the full anti-neoplastic effects of metformin.⁵⁷ Other studies defined metformin use as those with more than one prescription for metformin during the study period.^{1,57,58} However, these studies are limited by having a significant number of participants with low total number of prescriptions filed. Therefore, these subjects may not receive the full antineoplastic effects of metformin, which may contribute to the heterogeneity amongst the literature of metformin use and prostate cancer risk.

Another study similarly defined metformin exposure by subjects receiving at least one prescription.⁴ However, this study categorized subjects who were ever exposed to metformin according to the number of prescriptions received between cohort entry and up to 1 year prior to the index date.⁴ Additionally, this study excluded metformin users with under one year of metformin use. Therefore, this study was able to account for frequency of metformin use, while assessing a minimum of metformin use of at least one year, which allowed this study to better appreciate metformin's anti-neoplastic effects amongst the cohorts. Our proposed study will also utilize a 1-year minimum duration of metformin use, with medication duration as defined by the study database, which will be further detailed in chapter 3 of this paper.

2.4.4 Metformin dose

When examining the dose of metformin used in the literature, many studies had similar definitions of high and low doses of metformin. One study defined a high metformin dose as 1000mg or more per day. This was similar to a case control study that divided groups into a high dose and low dose group with a cut-off of 1g of metformin per day.⁵⁸ Another study standardized the amount of drug use by dividing the yearly amount of drug purchases by the average daily amount corresponding to a defined daily dose (DDD) specified by the WHO.⁵⁵ Therefore, the average yearly dosage was calculated by dividing the cumulative DDD amount by total years of use. Therefore, in analyses amongst high and low doses of metformin, 1 gram per day should be used as the cutoff point between high and low doses of metformin when examining medication dose as a study variable. Given the variability in the literature amongst metformin dose, our study will not separately analyze for a dose-dependent relationship of medication use and prostate cancer risk.

2.4.5 Metformin duration

As prostate cancer is a slow-growing disease, medication exposures prior to development may have an impact on disease development. The influence of duration of metformin use on the risk of prostate cancer is an important variable to consider in this retrospective cohort study. Additionally, there is substantial heterogeneity amongst studies in assessing the impact of duration of metformin usage on the risk of prostate cancer. For instance, one study determined an increase in prostate cancer risk with a greater number of metformin prescription acquired (RR 1.23; 95% CI 0.99-1.52; $P < 0.05$).⁴ This study included participants who ever used metformin during the study period.⁴ This finding contradicts other in vitro and in vivo studies that have demonstrated a decreased incidence of prostate cancer amongst metformin users.¹⁻³ However, the finding of an increased risk of prostate cancer among increased metformin durations was replicated by a study that examined both metformin and statins on prostate cancer risk, in which a longer proportion of time on a metformin daily dose at or above 1,000mg was associated with an increased prostate cancer incidence (HR 0.69; 95% CI 0.5-0.92; $P < 0.001$).⁵⁶ Interestingly, these findings contradict a sub-analysis on the effects of duration of metformin use on cancer risk, and only included patients on metformin for at least 365 days.¹ The researchers determined that one year of metformin use lowered the incidence of prostate cancer (HR 0.90; 95% CI 0.88-0.91; $P < 0.001$).¹ Another sub-analysis from a retrospective cohort study determined that incidence of prostate cancer decreased with cumulative duration of metformin use, and use above 44 months had the lowest incidence rate amongst the duration groups (HR 0.74; 95% CI 0.69-0.78; $P < 0.001$).⁵⁷ Therefore, studies varied widely in their duration of metformin usage, and the impact of the duration of metformin on the risk of prostate cancer remains unidentified.

2.4.6 Defining Statin Use

Understanding how statin-use is defined in prior studies will contextualize the findings amongst the literature and will support our study's definition of statin-use. There are different definitions of statin-use amongst the literature, however some studies defined statin use as at least one redeemed prescription 6-12 months before the index date.^{35,56} Conversely, in these studies, those redeeming their first statin prescription within 6 months prior to the index date were categorized as non-users.³⁵ This study design ensures that statin users are utilizing the medication for at least 6 months, therefore allowing the subjects to better receive the potential long-term anti-neoplastic effects of statins.

It is important to avoid including subjects who utilize statins for extended periods of time prior to study entry. This is necessary because those with extended statin use prior to cohort entry may receive anti-neoplastic effects that are not accounted for in analyses, therefore skewing the inverse relationship between statin use and prostate cancer incidence. To control for this, a study utilized a new-user design was adopted, in which patients who used a statin 180 days before cohort entry date were excluded, and further defined statin users as subject with at least one statin prescribed during the follow up period.⁵⁹ Based upon the literature, our study will define statin users as at least 1 year of medication use, as defined by prescriptions filled within our database, while limiting medication usage prior to cohort entry.

2.4.7 Statin dose

Unlike the relative homogeneity found amongst metformin dosing, the proper dose of statin for prevention of cancer has varied amongst several studies. One study defined a high dose of statin as the equivalent dose that reduces LDL by 30% or more on average.³¹ Another study categorized statins into three intensity groups- high, moderate, and low based on published guidelines and observational studies.⁵⁹ Both of these studies give evidence of a dose dependent

relationship of statins and prostate cancer,^{31,59} therefore it is crucial to consider the dose of statin used when assessing the incidence of prostate cancer amongst statin users. However, our study will not explore a dose-dependent relationship among statin use and prostate cancer risk due to the lack of consistent findings of a dose response of statin use and prostate cancer.

2.4.8 Statin duration

The association between duration of statin use and incidence of prostate cancer has been examined by several studies. One determined that a longer proportion of time on a higher statin daily dose was associated with a somewhat decreased prostate cancer incidence (HR 0.69; 95% CI 0.50-0.92; $P < 0.0001$).⁵⁶ Similarly, another found that cumulative duration of statin use over 12 months had significant reduction prostate cancer (HR 0.77; 95% CI 0.64-0.92; $P < 0.05$) when compared to statin use of 1-10 months (HR 1.88; 95% CI 1.63-2.17; $P < 0.05$).²⁷ Additionally a prospective cohort study found a substantial reduction in the risk of developing lethal prostate cancer among statin users, particularly subjects using statins longer than 5 years (HR 0.65; 95% CI, 0.47-0.90; $P < 0.05$).¹⁹ Interestingly, a case control study identified duration of statin use as independent variables of 0-2, 2-4, 5-9, or 10+ years and found that while the risk estimates did not differ substantially by duration of statin used, the inverse association between statin use and risk of prostate cancer was most pronounced amongst men who had used statins for more than 10 years (aOR 0.78; 95% CI 0.65-0.95; $P < 0.05$).³⁵

Conversely, a study that followed subjects for 7 years found no association amongst statin use and prostate cancer risk (HR 1.03; 95% CI 0.82-1.30; $P < 0.05$),²⁹ however this study did not stratify participants amongst total duration of statin usage. The discrepancies between studies on the effects of statins and prostate cancer incidence may be due to the duration studied. Very few studies have allowed for the long latency of any potential protective effects of statins, thus the follow up periods of many studies showing no association have been too short to detect

an impact.⁶¹ However, a meta-analysis of 15 retrospective studies that reported statin exposure for 5 years or more reported no impact on overall prostate cancer diagnosis (RR 0.89, 95% CI 0.66-1.12; $P < 0.05$), despite longer durations (5+ years) of statin use were associated a reduction in the risk of advanced prostate cancer.⁶¹ This suggests that longer durations of statin use may not be associated with alterations in prostate cancer incidence,²¹ which is contradictory to the above mentioned findings. Therefore, adjustments for statin duration are critical to implement given the inconsistencies amongst studies on the effects of statin duration on the incidence of prostate cancer.

2.4.9 Defining combination treatment of metformin and statins

There are a small number of studies that have examined the synergy of metformin and statin on the incidence of prostate cancer. One study examining the effect of statin use on prostate cancer risk in individuals already diagnosed with HGPIN and performed a sub analysis of several different medication classes and the risk of prostate cancer. They found that controlling for metformin did not alter the association between statins and a lower risk of prostate cancer (OR 0.45; 95% CI 0.21-0.97; $P < 0.04$).²² Only one other study examined the effects of both metformin and statins on prostate cancer risk, which was a 7-year cohort study within the VA health care system.⁵⁶ This study found that metformin was associated with a significantly reduced prostate cancer incidence among patients on statins (HR 0.69; 95% CI 0.50-0.92; $P < 0.0001$), while a comparatively increased prostate cancer incidence was found in those using metformin without statins (HR 2.15; 95% CI 1.83-2.52; $P < 0.05$).⁵⁶ Additionally there was a substantial reduction of prostate cancer incidence for those taking metformin and statins against those taking neither (HR 0.32; 95% CI 0.25-0.42; $P < 0.05$).⁵⁶ Therefore, despite the limited evidence on the combination of metformin and statins on the risk of prostate cancer, there is promising data of the potential beneficial effect of combination therapies.

2.4.10 Study Setting

As all participants will be males over the age of 50 with a confirmed diagnosis of BPH and T2DM, the most feasible option will be to analyze the population with a large database. Given the population being studied and need for detailed baseline information, the most appropriate study setting will be within the Veterans Administration (VA) health system database. Inpatient and outpatient settings will be included in the study setting, as a previous VA study utilized both inpatient and outpatient visits for classification of exposure.³⁶ All patients that will be enrolled will be prescribed in both inpatient and outpatient settings within the VA.

2.4.11 Confirmation of prostate cancer

When analyzing the incidence of prostate cancer in a population, it is crucial to have a standardized formulation to confirm prostate cancer. Information on subject's prostate cancer diagnoses will be confirmed from the VA database, using the ICD code for prostate cancer, "ICD-9-CM = 185". This method will be utilized because all patients registered in this database have been reviewed carefully to provide accurate diagnoses. As outlined by Kuo, et al., requirements for a confirmed code of prostate cancer must have sufficient evidence supporting their cancer diagnosis, such as histological or pathological reports, laboratory evidence or clinical images.³ Therefore, by utilizing the VA database, we can confirm an accurate diagnosis of prostate cancer in the subjects included into our study.

It is important to exclude those with undiagnosed underlying prostate cancer into the study. Therefore, a study excluded all prostate cancers diagnosed within 3 years after the initial baseline cohort entry to minimize latent prostate cancer at the baseline.⁵⁵ This same approach will be incorporated into our study to limit bias.

Some studies have utilized prostate-specific antigen to indirectly measure the development or progression of prostate cancer.⁶² However, prostate specific antigen may not be

an accurate measurement of prostate cancer because it is influenced by several extraneous variables.⁶³ By utilizing a retrospective cohort study with all included subjects already having a diagnosis of prostate cancer, we can eliminate the inaccuracy of utilizing prostate specific antigen to measure prostate cancer development.

2.4.12 Selection criteria

The selection criteria for studies completing a cohort study on the use of medications to reduce the risk of developing prostate cancer utilized various databases, with differing inclusion and exclusion criteria. For instance, studies utilized different age restrictions for inclusion criteria. One study utilized a subject population from the Veteran Health Administration Health Care System, and included subjects aged 18 to 90.⁵⁶ To confirm a proper diabetes diagnosis, participants had to have a diagnosis of T2DM (ICD- CM code of 250.00 or 250.02).⁵⁶ Another study included male patients aged at least 40 at the time of their first prescription of metformin.⁴ Similarly, another study selected males aged 50 years and older.³ Whereas another study restricted subjects ages to be 55-67 year old men.⁵⁵ Conversely, another study included all patients over 18 years. Therefore, there is wide variability amongst the selection criteria in studies that are examining prostate cancer incidence. Detailed inclusion and exclusion criteria will be included in chapter 3.

2.4.13 Follow up

There was considerable heterogeneity amongst the studies in their follow up period when analyzing medications and the risk of developing prostate cancer. The longest study follow up period was in a nested case-control study that included those who were first prescribed any oral hypoglycemic agent between 1988 and 2009, for a potential 22 year study period, however this study allowed a minimum of a 1 year follow up period.⁴ Therefore, subjects had a large variety in their exposure time. Another study included participants who initiated metformin between the

years 1999 and 2005, for a 6-year entrance period.⁵⁶ This study had a mean follow up of about 5 years, however the study was designed to follow participants for a maximum of 7 years.⁵⁶ Similarly, a prospective cohort study on the development of prostate cancer in statin users utilized a follow up period of only 7 years.²⁹ Another retrospective cohort study utilized a minimum of 180 days in the database as their minimum follow up criteria.⁵⁷ However, relatively short follow up periods may preclude evidence for a protective effects of medication if prostate cancers have not reached clinical detection by the end of the study.²⁶ This short follow up period was noted in several studies.^{35,36} Therefore, following patients for up to 10 years with a minimum of one year inclusion in the study period would provide an adequate amount of time for subjects to develop prostate cancer.

2.4.14 End point

Many of the studies on prostate cancer development utilize similar end points. These end points tended to be: first ever diagnosis of prostate cancer, death before a prostate cancer diagnosis during the study period, end of registration with the database, or end of the study period.^{4,55,56} Another study held similar end points, however the researches also added the last day of use of an oral glucose lowering drug, start of insulin or another oral glucose lowering drug other than metformin or sulfonylurea derivatives.¹ In studies examining the progression, rather than development, of prostate cancer tended to utilize other primary endpoints, such as Gleason scores.¹²

2.4.15 Statistical analysis

A majority of studies that examined the effect of metformin, statin, or a combination of the two on the risk of developing prostate cancer utilized either a logistic regression analysis,^{4,11} a cox proportional hazards model,^{19,26,27,29,37,55} or a combination of both analyses.¹² One study detailed that the cox proportional hazards model propensity score of medication group

membership was assigned to metformin and statin use.⁵⁶ These researchers described that this score is used to weight individuals differently to achieve balance in covariates at baseline between the four medication groups (metformin + no statin, metformin + statin, no metformin + no statin, no metformin + statin).⁵⁶ In a case control study determining the effect of metformin on the incidence of prostate cancer, the researchers utilized a conditional logistic regression analysis.⁴ In an examination of metformin against sulfonylureas in risk of prostate cancer, statistical analyses were performed with a cox proportional hazard model with cumulative duration of drug use as a time varying determinant.¹ Similar methods of using drug exposure as a time dependent variable were completed in studies analyzing statin use on incidence of prostate cancer.²⁹ Performing statistical analyses with a time dependent variable allows researchers to avoid immortal time biases.

Another crucial bias to avoid in cox proportional hazard models are survival bias. In one study, survival bias was avoided by modeling all cumulative drug exposures after prostate cancer as time dependent covariates.⁸ Therefore, the comparison is not exclusively between users and nonusers but is also between users who have had different durations of exposure.⁸ By considering medications as time dependent variables, those ever using metformin contribute person-years to their respective group.⁵⁷ Doing so allows for statistical consideration of duration of metformin use as a variable in the analysis. Therefore, it will be critical to avoid immortal time biases and survival biases when performing cox proportional hazards models.

2.4.16 Statistical significance

The majority of the literature on the effect of metformin or statins on prostate cancer determined statistical significance using an alpha of 0.05 and power of 80%.^{2,55-57} Our study will utilize the same alpha and power to determine statistical significance and calculate our sample size, with further details in chapter 3 and Appendix B.

2.4.17 Sample size

When examining cohort and case-control studies, the sample sizes of the study population tend to be large. As the literature review did not contain any randomized control trials determining the effect of metformin or statins on prostate cancer incidence, our sample size calculation included data from observational studies. Using two studies in the literature, we calculated the effect size for our population and various cohorts.

First, Kuo et al. conducted a retrospective cohort study evaluating metformin use on the risk of prostate cancer in subjects with BPH.³ The researchers measured the effect of metformin use with the number of prostate cancer diagnoses within the study timeline. After proper exclusion, the metformin and the non-metformin cohort each contained 2906 participants. After 10 years, the number of prostate cancer diagnoses within the metformin cohort was 72, whereas the number of prostate cancer diagnoses within the non-metformin cohort was 94. This study was chosen to represent the mean expected prostate cancer incidence amongst our metformin only cohort.

Second, Lehman et al. utilized a prospective cohort study to determine the influence of statin therapy amongst the incidence of prostate cancer in metformin users.⁵⁶ This study also measured the effect of medication use through the number of prostate cancer diagnoses within the study timeframe. The non-metformin, non-statin cohort had 1930 participants, with 186 incident prostate cancer diagnoses with an average follow up time of 1,739.77 days +/- 908.86 days. The non-metformin, statin user cohort had 2404 participants, with 135 prostate cancer diagnoses with an average follow up time of 2080.83 days +/- 825.99 days. The metformin, non-statin cohort had 175 participants, with 22 prostate cancer diagnoses with an average follow up time of 1,706.44 days +/- 932.53 days. Lastly, the metformin and statin cohort had 533 participants, with 17 prostate cancer diagnoses with an average follow up time of 1,766.37 days

+/- 829.96 days. This study was chosen to represent the comparison group, as well as the mean expected prostate cancer incidence for the metformin + statin cohort and statin only cohort.

The information provided in the above studies will allow for proper calculations of our study's sample size, which is detailed in chapter 3 of this paper.

Interestingly, the heterogenous results on statin and prostate cancer incidence can be related to differences in sample sizes. Small study populations in some studies⁶⁴ invite subjectivity and may lack crucial data. When examining the two largest cohorts amongst statin studies, Murtola showed a 7% increase in risk (HR 1.42; 95% CI 1.04-1.95; $P < 0.05$)⁴² while Jespersen showed a 6% decrease in risk (aOR 0.78; 95% CI 0.65-0.95; $P < 0.05$)³⁵ of prostate cancer amongst statin users. However, neither estimate was adjusted for PSA screening. These findings demonstrate the importance of a large sample size with adequate controls for confounders, which will be achievable using the VA database.

2.5 Conclusion

The information presented in this literature review chapter details the necessity of the proposed study. There is evidence that BPH is associated with prostate cancer, and that combined treatment of metformin and statins may reduce the incidence of prostate cancer. The above chapter also highlights the heterogeneity amongst the findings, which can be partly explained by confounding variables. This study will determine if statins and metformin can be utilized as a primary prevention modality to reduce the risk of developing prostate cancer in individuals with T2DM and BPH. The study that will be detailed in chapter 3 will examine a method to reduce the risk of developing one of the most common causes of cancer related mortality amongst men and therefore can assist with keeping the aging population healthy.

2.6 References

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Chapter 3: Study Methodology

3.1 Study design

The proposed study design will be a retrospective cohort study within the Department of Veterans Affairs that will investigate the association between statin use, metformin use, and the incidence of prostate cancer in men over the age of 50 with BPH and T2DM who utilized either metformin alone, statins alone, or a combination of metformin and statins for at least 1 year during the years 2009-2019. As this will be a retrospective study, participants and researchers will not be blinded, and participants will not be notified of their inclusion in the study. Data will be sourced from the Veterans Affairs and the Catastrophic Patient Database for confirmation of benign prostatic hyperplasia, type 2 diabetes mellitus, and prostate cancer by using ICD codes. No identifiable patient information will be shared other than to match Catastrophic Patient Database records to VA records.

The study design will be a retrospective cohort study, as those who would eventually develop prostate cancer during the follow up period would already have this outcome of interest prior to the start of this study. While a randomized control trial would provide significant insight into the relationship between statins, metformin, and the incidence of prostate cancer amongst men with T2DM and BPH, it is not feasible to complete this study within the allotted two-year time frame due to the slow growing nature of cancerous prostate cells. Therefore, this study will utilize a retrospective cohort approach, with a 10 year follow up period. As explained in chapter 2, a 10 year follow up allows for a proper length of time to develop prostate cancer amongst at risk individuals. Subjects who fit the inclusion criteria will enter the study cohort in the year 2009 and will be followed until the year 2019. New subjects may not be added into the study group after the year 2009. Subjects must be followed up for at least 1 year. The VA database will

be utilized for study population selection. Of those within the VA database who fit the inclusion criteria, subjects will be assigned into cohorts based upon their exposure status. Participants will be divided into four cohorts: statin and metformin treatment, statin alone treatment, metformin alone treatment, and no statin or metformin treatment. These cohort groups are reproduced in Table 1 and Appendix A. Cohorts will be matched by age, propensity score for comorbidities, and index date of benign prostatic hyperplasia and diabetes diagnoses. Those in cohort group 4 (those who did not utilize statins and utilized a non-metformin oral hypoglycemic agent) will serve as the unexposed cohort. The endpoint of follow-up for the study population will be the date of prostate cancer diagnosis, death, termination of enrollment from the selected database, or the end of the follow up period.

Table 1: Baseline Demographic and Clinical Characteristics of Subjects

	Metformin + Statin	Metformin Only	Statin Only	Non metformin + Non statin	P value
Demographics					
Age, y	Mean (sd)	Mean (sd)	Mean (sd)	Mean (sd)	t-test
Age Group					
50-64	Mean (sd)	Mean (sd)	Mean (sd)	Mean (sd)	t-test
65- 79	Mean (sd)	Mean (sd)	Mean (sd)	Mean (sd)	t-test
80+	Mean (sd)	Mean (sd)	Mean (sd)	Mean (sd)	t-test
Race					
White	(%)	(%)	(%)	(%)	Chi square
Black	(%)	(%)	(%)	(%)	Chi square
Hispanic Ethnicity	(%)	(%)	(%)	(%)	Chi square
Other	(%)	(%)	(%)	(%)	Chi square
PCa Screening					
PSA (times/year)	Mean (sd)	Mean (sd)	Mean (sd)	Mean (sd)	t-test
Comorbidities					
Myocardial Infarction	(%)	(%)	(%)	(%)	Chi square
Congestive heart failure	(%)	(%)	(%)	(%)	Chi square
Peripheral vascular disease	(%)	(%)	(%)	(%)	Chi square
Cerebrovascular disease	(%)	(%)	(%)	(%)	Chi square
Dementia	(%)	(%)	(%)	(%)	Chi square
Chronic lung disease	(%)	(%)	(%)	(%)	Chi square
Ulcer	(%)	(%)	(%)	(%)	Chi square
Chronic liver disease	(%)	(%)	(%)	(%)	Chi square
Hemiplegia	(%)	(%)	(%)	(%)	Chi square
Kidney disease	(%)	(%)	(%)	(%)	Chi square
Tumor, leukemia, lymphoma	(%)	(%)	(%)	(%)	Chi square
Moderate or severe liver disease	(%)	(%)	(%)	(%)	Chi square
Malignant tumor, metastasis	(%)	(%)	(%)	(%)	Chi square
AIDS	(%)	(%)	(%)	(%)	Chi square
Retinopathy	(%)	(%)	(%)	(%)	Chi square
Nephropathy	(%)	(%)	(%)	(%)	Chi square
Neuropathy	(%)	(%)	(%)	(%)	Chi square
Erectile Dysfunction	(%)	(%)	(%)	(%)	Chi square
Prostate cancer	(%)	(%)	(%)	(%)	Chi square
Charlson Comorbidity Index					
0	(%)	(%)	(%)	(%)	Chi square
1	(%)	(%)	(%)	(%)	Chi square
2+	(%)	(%)	(%)	(%)	Chi square

3.2 Study population and sampling

The study population will be individuals at a baseline heightened incidence rate of prostate cancer. As both increased age and BPH are associated with increased incidence of prostate cancer, the study population will include those over the age of 50 with BPH and T2DM. These diseases will be confirmed using ICD-CM codes (T2DM = 250.X and BPH = 600.X). Prostate cancer will be validated by requiring subjects to have their diagnosis identified by the database. As prostate cancer takes an extended amount of time to develop, subjects will be followed for a 10-year duration from 2009-2019, with a minimum of 1 year follow up duration. Recruitment into the study will not require any notification to the participants as this is a retrospective study, thus participants will not be exposed to an intervention. Sampling for the study will be conducted through analyses within the VA database, such as the Corporate Data Warehouse (CDW) and evaluated through the Capri, Reach Vet, Risk Indicators and Storm Tool for Analytic Look Up (CRISTAL) as well at the PBM v3.0 Database which are further detailed in this chapter.

3.3 Inclusion criteria

The inclusion criteria of our study are that subjects, at baseline, are males aged 50 years or older with confirmed diagnoses of BPH and T2DM. Subjects must be followed for at least 1 year. Subjects classified as metformin users must have regular metformin usage of at least 6 months, as defined by a continuous prescription of metformin for 6 months within 1 year. Subjects classified as statin users must have regular statin usage of at least 6 months, as defined by a continuous prescription of statins for 6 months within 1 year. Subjects in the unexposed cohort may utilize any non-metformin hypoglycemic agent, such as: sulfonylureas, SGLT-2 inhibitors, GLP-1 receptors antagonists, thiazolidinediones, etc.

3.4 Exclusion criteria

Those without database confirmed diagnoses of BPH or T2DM will be excluded from the study. Those under the age of 50 during the year 2009 will be excluded from the study. Those without metformin or statin usage for at least 6 months within 1 year will be excluded from the study. If subjects switch from metformin to a non-metformin hypoglycemic agent during the study period, they will be excluded from the study. Those with exposure to metformin or statins in the year immediately prior to the index date (medication initiation between January 1st 2008 – December 31st 2008) will be excluded to take into account the long latency period of prostate cancer and to minimize detection bias, where initiation of a new treatment may lead to an increased intensity of diagnostic investigations, possibly leading to an increased probability of detecting cancer.¹ Additionally, subjects utilizing 5-alpha reductase inhibitors will be excluded due to their potential effect to lower the incidence of low-grade prostate cancer.

3.5 Subject protection and confidentiality

The Yale University Human Investigation Committee and the Yale Human Research Program must approve all aspects of this study and will require records throughout the study. All researchers involved in the study will be HIPPA certified prior to commencement of the study. All electronic patient data will be maintained on a secure, encrypted platform on VA computer systems that require access with multifactor authentication. All patients will be de-identified by removing patient identification, and patients will be assigned a random number for identification. Additionally, the Department of Veterans Affairs Central IRB requires that all principal investigators involved in the study to complete VA central IRB form 108 for data to be accessed for research. The VA will have a local site investigator located at the West Haven VA, who will ensure that all patient data is properly controlled. Finally, study approval of the West Haven VA

IRB will be required for data access and storage. The IRB approval process is detailed in Appendix C.

3.6 Recruitment

Participants will be recruited by utilizing various aspects within the VA and VHA. First, the Corporate Data Warehouse (CDW) will be used to design the initial list of potential participants. The CDW database is updated every day and contains all electronic health records within the VHA. Subjects will be further analyzed using the Capri, Reach Vet, Risk Indicators and Storm Tool for Analytic Look Up (CRISTAL). This tool uses CDW data to compile databases that are accessible for studies without needing input from an investigator. Using CRISTAL, we will compile a list of veterans who have received VHA care and are diagnosed with BPH and T2DM. Subjects will be cross referenced with the PBM v3.0 Database to identify participants that have utilized either metformin, statins, or both. The PBM v3.0 Database contains extensive details on the medications prescribed and the characteristics of the prescriptions (i.e. days supplied). As the PBM database is not available at the CDW, extract requests will be sent through the PBM intranet site. The PBM v3.0 database will be used to separate the cohorts into the exposure groups (metformin only, statin only, and metformin + statin combination) and the comparison group (no metformin + no statin). Using CRISTAL, the curated list of veterans with type 2 diabetes mellitus and benign prostatic hyperplasia who have received VHA care will be identified, and CRISTAL will flag participants that received metformin or statin prescriptions. This final list will comprise the study cohort.

3.7 Study variables and measures

The independent variables will be treatment with either metformin, statins, or both medications. The dependent variable is prostate cancer diagnoses amongst the cohorts.

3.8 Methodology considerations

In this retrospective cohort study, the main exposure will be receiving either a metformin, statin, or combination prescription for at least 1 year during the years 2009-2019. The comparison will be participants who have not received either metformin or statin during the study timeline.

The primary outcome is the incidence of prostate cancer diagnoses amongst the cohorts. This will be a dichotomous variable within groups.

3.9 Confounding variables

This study will account for many of the confounding variables described in chapter 2. Confounders will be matched amongst those in the comparison and exposure groups in several categories. The first category will match comparison and control categories by various demographics, including age, race, and BMI. The second category will match categories by health characteristics, including average cholesterol levels, PSA levels, fasting blood sugar levels. Health characteristics will be controlled using the Charlson comorbidity index, which is an index used to predict ten-year survivability. All confounders and treatment groups are summarized in table 1. Statistical analyses will be performed between exposure and treatment groups to examine the primary outcome.

3.10 Data collection

Methods for data collection is outlined in section 3.3 under study recruitment. The CDW will be used to gather an initial list of potential participants. This data will be further specified using CRISTAL and PMB v3.0.

3.11 Sample size calculation

The sample size was calculated from two studies with the primary outcome of incidence of new prostate cancer diagnoses, a dichotomous variable, in metformin or statin users. The sample size was first calculated using STATCALC, which is the Centers for Disease Control and

Prevention sample size software. This software was utilized because of its ability to calculate a minimum required number of subjects needed for a retrospective cohort study. Additionally, this software utilizes several sample size calculation formulas, one of which is the Fleiss with Correction for Continuity. This formula prevents under-estimation of sample sizes, which can be observed with uncorrected formulas for sample size in retrospective cohort studies. Then, the G power 2 calculator was utilized to ensure the accuracy of this calculation. This calculation was made on the assumption that a dichotomous variable would be compared between the cohorts and comparison group.

The mean of the population studied was extrapolated from Kuo, et al.² and Lehman, et al.³ Based on Kuo, et. al, the clinically significant difference for metformin-only users to be expected would be an adjusted hazard ratio of 0.69 (95% CI .49-.96; p=0.0039),² with an estimated effect size for our metformin-only cohort of .75%. Based on Lehman et. al, the clinically significant difference for statin-only users to be expected would be an adjusted hazard ratio of 0.60 (95% CI .502-.709; P <0.0001) with an estimated effect size for the statin-only cohort of 4.02%.³ Lastly, according to Lehman et. al, the clinically significant difference for statin and metformin users would be an adjusted hazard ratio of 0.31 (95% CI 0.242-0.406; P <0.0001) with an estimated effect size for the metformin + statin cohort of 6.45%.³ These hazard ratios and effect sizes were utilized to calculate sample size through the Fleiss with Correction for Continuity formula.

This study will account for a loss to follow up, as a Kuo et. al had 2% of participants that were followed for less than 1 year. Utilizing the Fleiss formula with correction for continuity, and assuming a 1:1 ratio of exposed to unexposed, there will need to be a minimum of 17,323 total patients evaluated in this study, 256 in the statin and metformin cohort, 733 in the statin

only cohort, 8,167 in the metformin only cohort, and 8167 in the non-statin and non-metformin cohort to power this study at 80% with an alpha of 0.05. As there were approximately 6,000,000 patients seen per year in VA facilities during the years 2011 and 2018,³ our sample size will be attainable within the VA database. A summary of the sample size calculations is provided in Appendix B.

3.12 Analysis

Statistical Analysis System Software (SASS) will be used to analyze all data collected by CDW and other VA databases. Baseline characteristics that may serve as confounders will be compared through Chi-Squared tests (table 1). The primary outcome of this study will be analyzed using the cox proportional hazards regression analysis to determine hazard ratios of the cohorts. Medication use will be treated as a time-dependent covariate. The Kaplan Meier method will be used to determine the cumulative incidence of prostate cancer in the cohorts, and the difference between cohorts will be tested with the log rank test. Sensitivity analyses will be performed using the Cox regression hazards model on subgroups classified by comorbidity. These statistical analyses are demonstrated in Table 2 below.

Table 2: Incidence of Prostate Cancer by Metformin and/or Statin Exposure

	Case number observed	Incident cases of prostate cancer	Person years	Incidence rate (per 100,000 person-years)	Hazard Ratio
Metformin + Statin Cohort					
<i>Metformin cumulative duration (months)</i>					
6-12 mth	N	N	N person years	N	HR (95% CI)
13-24 mth	N	N	N person years	N	HR (95% CI)
25+ mth	N	N	N person years	N	HR (95% CI)
<i>Statin cumulative duration (months)</i>					
6-12 mth	N	N	N person years	N	HR (95% CI)
13-24 mth	N	N	N person years	N	HR (95% CI)
25+ mth	N	N	N person years	N	HR (95% CI)
Metformin Only Cohort					
<i>Metformin cumulative duration (months)</i>					
6-12 mth	N	N	N person years	N	HR (95% CI)
13-24 mth	N	N	N person years	N	HR (95% CI)
25+ mth	N	N	N person years	N	HR (95% CI)
Statin Only Cohort					
<i>Statin cumulative duration (months)</i>					
6-12 mth	N	N	N person years	N	HR (95% CI)
13-24 mth	N	N	N person years	N	HR (95% CI)
25+ mth	N	N	N person years	N	HR (95% CI)

3.13 Timeline and resources

This study is estimated to take place over six months. The first two months will encompass collecting data from the VA databases. The following two months will be allotted for analysis of the data. The final two months will be for extrapolating results and finalizing a paper for publication. This study requires access to the various VA databases through the West Haven VA. The primary investigator will be Dr. Harris Foster, MD, and the co-primary investigator will be Kevin Mugno, PA-SII.

3.14 References

1. Azoulay L, Dell'Aniello S, Gagnon B, Pollak M, Suissa S. Metformin and the incidence of prostate cancer in patients with type 2 diabetes. *Cancer Epidemiology Biomarkers and Prevention*. 2011;20(2):337-344.
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Chapter 4: Conclusion

4.1 Advantages and Disadvantages

Given the novelty and clinical importance of this study, it has the advantage of significantly impacting the health outcomes of those at risk for one of the most common cancers amongst elderly male populations. Additionally, this study has many advantages due to its scope, design, and methodology. One advantage of the proposed study is that it will utilize data that is collected through VA databases. Being able to access this database will significantly lower the time, cost, and resources required to conduct this type of study. Another advantage of this study is its generalizability. Utilizing national VA databases that encompasses all VA health records will allow the study population to accurately represent the general population of men with diabetes and BPH. This study can be generalized to the non-veteran population because the same statin and metformin usage and risk factors for prostate cancer exist within the non-veteran population. A final advantage of this study is that numerous confounder variables will be accounted for by matching between cohorts. Matching among participants in this retrospective study will be more accurate than a prospective study because the recruitment process in a retrospective study can be more exhaustive due to increased baseline data.

However, this study contains several disadvantages. One of the most significant disadvantages is due to the retrospective design of the study, as inferences about causality amongst the relationship between metformin or statin use and prostate cancer risk among individuals with benign prostatic hyperplasia cannot be made. Therefore, the only inference that can be made from this study is a potential statistically significant relationship among statin and/or metformin use and prostate cancer incidence among individuals with benign prostatic hyperplasia. Another disadvantage from this retrospective design of this study is that all subjects

are assumed to be fully adherent to their medication regimens. This assumption may falsely skew the results of the study, as non-adherent subjects may be present amongst the various medication cohorts. A final disadvantage of this study is the inability to control for duration of metformin and/or statin use. As individuals in the study design could utilize the medications for any time between 1-10 years, the potential duration dependent effect of this relationship may factor into the results of the study.

4.2 Feasibility

This study is feasible because all data needed for this study is included in the VA databases. The limitations to this study's feasibility would be IRB approval from the VA.

4.3 Mechanistic significance

As this study is a retrospective cohort study with an inability to control for all theoretical confounders, it contains low mechanistic significance.

4.4 Clinical significance

This study is clinically significant as it is both novel and of great clinical necessity. This study would be the first to examine the relationship of multiple medications that may lower the incidence of prostate cancer in those with benign prostatic hyperplasia. There remains a gap in the current literature amongst the relationship of metformin and statins in the risk of prostate cancer in high-risk populations. The results of this study will grow the knowledge of prostate cancer prevention. Further, these findings can aid in developing therapeutics to reduce the incidence of one of the most common cancers amongst male populations.

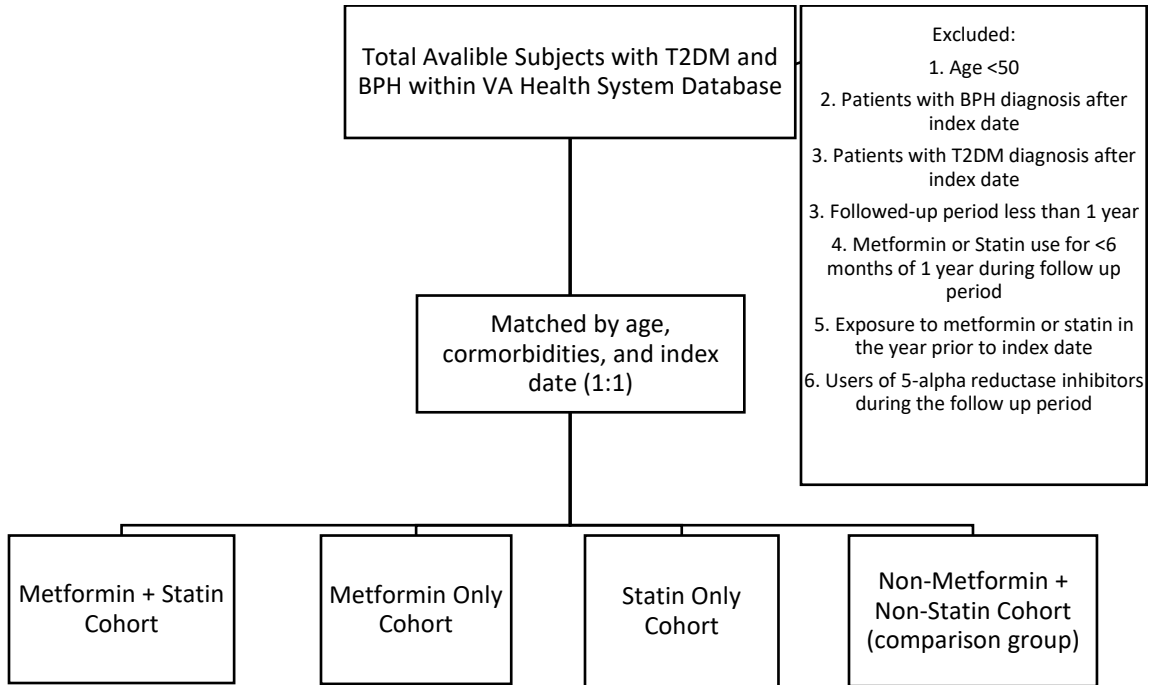
4.5 Future direction

If the hypothesized relationship exists, it will add evidence that may ultimately lead to the implementation of metformin and/or statins to lower the risk prostate cancer in high-risk populations. Depending on our conclusions, a future study may utilize a similar study population, exposure, and outcome with a randomized control trial with the intent to determine causality

among metformin and statin use and prostate cancer incidence. These findings could aid clinicians in prescribing appropriate medications for those assumed to be at an increased risk for developing prostate cancer.

Appendices

Appendix A: Study Profile



Appendix B: Sample Size Calculations

1. Sample size for Metformin + Statin cohort

StatCalc - Sample Size and Power

Unmatched Cohort and Cross-Sectional Studies (Exposed and Nonexposed)

Two-sided confidence level: 95%

Power: 80%

Ratio (Unexposed : Exposed): 1

% outcome in unexposed group: 9.64%

Risk ratio: 0.33092

Odds ratio: 0.30887

% outcome in exposed group: 3.19%

	Kelsey	Fleiss	Fleiss w/ CC
Exposed	227	226	256
Unexposed	227	226	256
Total	454	452	512

Effect size 6.45%

2. Sample size for Metformin-only cohort

StatCalc - Sample Size and Power

Unmatched Cohort and Cross-Sectional Studies (Exposed and Nonexposed)

Two-sided confidence level: 95%

Power: 80%

Ratio (Unexposed : Exposed): 1

% outcome in unexposed group: 3.23%

Risk ratio: 0.7678

Odds ratio: 0.7619

% outcome in exposed group: 2.48%

	Kelsey	Fleiss	Fleiss w/ CC
Exposed	7741	7739	8004
Unexposed	7741	7739	8004
Total	15482	15478	16008

Effect size 0.75%

3. Sample size for Statin-only cohort

StatCalc - Sample Size and Power

Unmatched Cohort and Cross-Sectional Studies (Exposed and Nonexposed)

Two-sided confidence level: 95%

Power: 80%

Ratio (Unexposed : Exposed): 1

% outcome in unexposed group: 9.64%

Risk ratio: 0.58299

Odds ratio: 0.55816

% outcome in exposed group: 5.62%

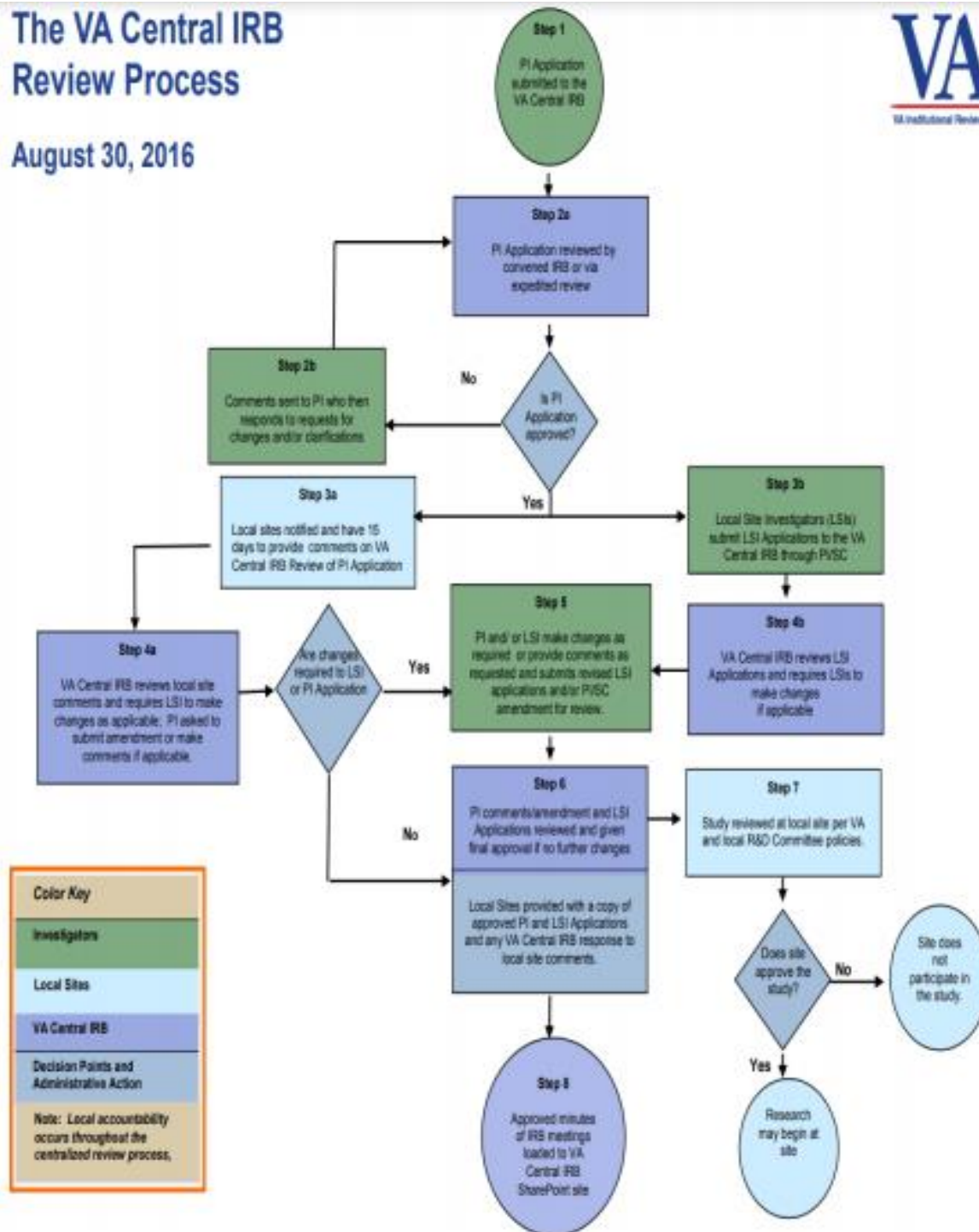
	Kelsey	Fleiss	Fleiss w/ CC
Exposed	685	684	733
Unexposed	685	684	733
Total	1370	1368	1466

Effect size 4.02%

Appendix C: VA IRB Approval Process

The VA Central IRB Review Process

August 30, 2016



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